

IN THE CLAIMS:

1. (Currently amended) A ~~phosphoprotein detection reagent (PPDR)~~ composition comprising a membrane having bound phosphoprotein, wherein the phosphoprotein is coordinated to a phosphoprotein detection reagent (PPDR) comprising:
  - (i) a polydentate chelator coordinated to a metal ion selected from the group consisting of  $\text{Fe}^{3+}$ ,  $\text{Al}^{3+}$ ,  $\text{Yb}^{3+}$ , and  $\text{Ga}^{3+}$ ; and
  - (ii) a detectable moiety conjugated to the polydentate chelator at a site other than a potential metal ion coordination site; and
  - ~~(iii) — a binding solution with a pH ranging from about 5.0 to about 7.0,~~ wherein the chelated metal ion selectively binds to a phosphorylated amino acid residue in a the phosphoprotein ~~if present~~ to create a chelator-metal ion-phosphoprotein (CMPP) complex; and the detectable moiety allows the CMPP complex to be detected ~~if present~~.
2. (Currently amended) The composition PPDR of claim 1, wherein the PPDR is soluble in an aqueous medium.
3. (Currently amended) The composition reagent of claim 1, wherein the chelator is a tetradentate nitriloacetic acid.
4. (Currently amended) The composition reagent of claim 1, wherein the chelator is a tridentate iminodiacetic acid.
5. (Canceled)
6. (Currently amended) The composition reagent of claim 1, wherein the metal ion is  $\text{Ga}^{3+}$ .
7. (Currently amended) The composition reagent of claim 1, wherein the metal ion is  $\text{Fe}^{3+}$ .

8. (Currently amended) The composition reagent of claim 1, wherein the detectable moiety is biotin.

9. (Currently amended) The composition reagent of claim 1, further comprising a spacer between the chelator-metal ion moiety and the detectable moiety.

10. (Currently amended) A method for synthesizing preparing a composition comprising a membrane having bound phosphoprotein, wherein the phosphoprotein is coordinated to a phosphoprotein detection reagent (PPDR), the method comprising:

- (a) reacting a polydentate chelator donor molecule with a detectable moiety donor under conditions wherein a detectable moiety is transferred to a polydentate chelator at a site other than a coordination site to form a chelator-detectable moiety complex; and
- (b) chelating a metal ion selected from the group consisting of  $\text{Fe}^{3+}$ ,  $\text{Al}^{3+}$ ,  $\text{Yb}^{3+}$ , and  $\text{Ga}^{3+}$  to the polydentate chelator to form a PPDR, wherein the PPDR is soluble in aqueous medium; and
- (c) contacting a membrane having bound phosphoprotein with the PPDR.

11. (Original) The method of claim 10, wherein the chelator donor molecule is selected from the group consisting of 2-(aminooxyethyl)iminodiacetic acid (AIDA), aminobutyl-nitriloacetic acid (AB-NTA), and iminodiacetic acid (IDA).

12. (Original) The method of claim 10, wherein the detectable moiety donor is selected from the group consisting of sulfo-N-hydroxysuccinimidyl-biotin (sulfo-NHS-biotin), sulfosuccinimidyl-6-(biotinamido) hexanoate (sulfo-NHS-LC-biotin), sulfosuccinimidyl-6-(biotinamido)-6-hexanimido hexanoate (sulfo-NHS-LC-LC-biotin), and penta-fluorophenyl-biotin.

13. (Original) The method of claim 10, wherein the detectable moiety donor is present in the reacting step in a molar excess over the polydentate chelator donor molecule.

14. (Previously presented) The method of claim 10, wherein the chelator-detectable moiety complex and a metal ion-containing solution are present in equimolar concentrations in the chelating step.

15-35. (Canceled)

36. (Currently amended) A kit comprising:

(a) a phosphoprotein detection reagent (PPDR) comprising:

(i) a polydentate chelator coordinated to a metal ion selected from the group consisting of  $\text{Fe}^{3+}$ ,  $\text{Al}^{3+}$ ,  $\text{Yb}^{3+}$ , and  $\text{Ga}^{3+}$ ; and

(ii) a detectable moiety conjugated to the polydentate chelator at a site other than a potential metal ion coordination site,

wherein the chelated metal ion selectively binds to a phosphorylated amino acid residue in a phosphoprotein if present to create a chelator-metal ion-phosphoprotein (CMPP) complex, and the detectable moiety allows the CMPP complex to be detected if present; and

(b) a membrane; and

(c) instructions for using the PPDR.

37. (Canceled)

38. (Original) The kit of claim 36, further comprising a secondary reagent for detecting the PPDR.

39. (Previously presented) The kit of claim 36, wherein the phosphoprotein detection reagent (PPDR) is soluble in aqueous medium.

40. (Canceled)

41. (Currently amended) A composition comprising a membrane having bound phosphoprotein, wherein the phosphoprotein is coordinated to a phosphoprotein

detection reagent (PPDR) ~~composition~~ comprising a chelator and a detectable moiety conjugated to the chelator in a binding solution with a pH ranging from about 5.0 to about 7.0, wherein:

- (i) the chelator comprises a tetradentate nitriloacetic acid or a tridentate iminodiacetic acid coordinated to a metal ion selected from the group consisting of  $\text{Fe}^{3+}$ ,  $\text{Al}^{3+}$ ,  $\text{Yb}^{3+}$ , and  $\text{Ga}^{3+}$ ;
- (ii) the chelated metal ion selectively binds to a phosphorylated amino acid residue in the ~~a~~-phosphoprotein ~~if present~~ to create a chelator-metal ion-phosphoprotein (CMPP) complex, and the detectable moiety allows the CMPP complex to be detected ~~if present~~; and
- (iii) the PPDR is soluble in aqueous medium.

42. (Currently amended) The composition ~~phosphoprotein detection reagent (PPDR)~~ of claim 41, wherein the metal ion is  $\text{Ga}^{3+}$ .

43. (Currently amended) The composition ~~phosphoprotein detection reagent (PPDR)~~ of claim 41, wherein the metal ion is  $\text{Fe}^{3+}$ .

44. (Currently amended) The composition ~~phosphoprotein detection reagent (PPDR)~~ of claim 41, wherein the detectable moiety is biotin.

45. (Currently amended) The composition ~~phosphoprotein detection reagent (PPDR)~~ of claim 41, further comprising a spacer between the chelator and the detectable moiety.

46. (Currently amended) A composition comprising:

- (a) a membrane;
- (b) a phosphoprotein bound to the membrane;
- (c) a metal ion selected from the group consisting of  $\text{Fe}^{3+}$ ,  $\text{Al}^{3+}$ ,  $\text{Yb}^{3+}$ , and  $\text{Ga}^{3+}$ ;

- (d)(b) a phosphoprotein detection reagent (PPDR) comprising a chelator and a detectable moiety, wherein:
- (i) the detectable moiety is conjugated to the chelator at a site other than a potential metal ion coordination site;
  - (ii) the chelator comprises a polydentate chelator coordinated to the metal ion to form a chelator-metal ion moiety;
  - (iii) the chelator-metal ion moiety selectively binds to a phosphorylated amino acid residue in the a phosphoprotein ~~if present~~ to create a chelator-metal ion-phosphoprotein (CMPP) complex; and
  - (iv) the detectable moiety allows the CMPP complex to be detected ~~if present~~; and
- (c) a binding solution having a pH ranging from about 5.0 to about 7.0, wherein the chelated metal ion selectively binds to the phosphorylated amino acid residue in the phosphoprotein, ~~if present~~, in the binding solution.

47. (Previously presented) The kit of claim 36, wherein the kit further comprises a binding solution having a pH ranging from about 5.0 to about 7.0.

Please add the following new claim:

48. (New) The composition of claim 1, comprising a binding solution with a pH ranging from about 5.0 to about 7.0